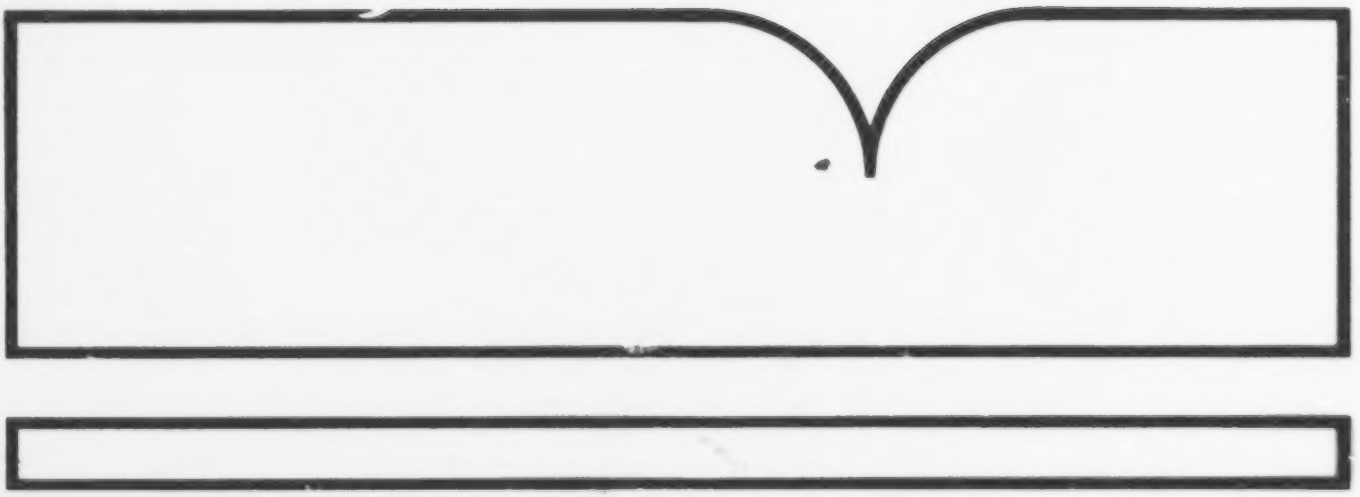
PB82-155995

The Determination of Chlorinated Herbicides in Industrial and Municipal Wastewater: Method 615

(U.S.) Environmental Monitoring and Support Lab.-Cincinnati, OH

Feb 82



U.S. Department of Commerce
National Technical Information Service
NTIS

PB82-155995

THE DETERMINATION OF CHLORINATED HERBICIDES IN INDUSTRIAL AND MUNICIPAL WASTEWATER

Method 615

Thomas A. Pressley
and
James E. Longbottom
Physical and Chemical Methods Branch
Environmental Monitoring and Support Laboratory
Cincinnati, Ohio 45268

January 1982

ENVIRONMENTAL MONITORING AND SUPPORT LABORATORY
OFFICE OF RESEARCH AND DEVELOPMENT
U.S. ENVIRONMENTAL PROTECTION AGENCY
CINCINNATI, OHIO 45268

REPRODUCED BY
NATIONAL TECHNICAL
INFORMATION SERVICE
U.S. DEPARTMENT OF COMMERCE
SPRINGFIELD, VA 22161

TECHNICAL REPORT DATA
(Please read Instructions on the reverse before completing)

1. REPORT NO.	2.	3. RECIPIENT'S ACCESSION NO.
EPA-600/4-82-005	REPORT	PB82 155995

4. TITLE AND SUBTITLE	5. REPORT DATE
The Determination of Chlorinated Herbicides in Industrial and Municipal Wastewater Method 615	February 1982
	6. PERFORMING ORGANIZATION CODE 033017

7. AUTHOR(S)	8. PERFORMING ORGANIZATION REPORT NO.
Thomas A. Pressley and James E. Longbottom	

9. PERFORMING ORGANIZATION NAME AND ADDRESS	10. PROGRAM ELEMENT NO.
Environmental Monitoring and Support Lab - Cincinnati Office of Research and Development U. S. Environmental Protection Agency Cincinnati, Ohio 45268	ABEB1C
	11. CONTRACT/GRANT NO.

12. SPONSORING AGENCY NAME AND ADDRESS	13. TYPE OF REPORT AND PERIOD COVERED
Same as Above	Final
	14. SPONSORING AGENCY CODE EPA/600/6

15. SUPPLEMENTARY NOTES
1 of a 12-method package

16. ABSTRACT

This is a gas chromatographic (GC) method applicable to the determination of selected pesticides in municipal and industrial discharges as provided under 40CFR 136.1. An acidified sample is solvent extracted with ethyl ether using a separatory funnel. Esters of the acids are hydrolyzed to the free acid. The acids are esterified with diazomethane and determined by GC using an electron capture or halide detector.

The compounds included in the method scope are the salts, esters, or acids of 2,4-D; 2,4-DB; 2,4,5-T; 2,4,5-TP; dalapon; dicamba; dichloroprop; dinoseb; MCPA; and MCPP.

17. KEY WORDS AND DOCUMENT ANALYSIS

a. DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group

18. DISTRIBUTION STATEMENT	19. SECURITY CLASS *(This Report)*	21. NO. OF PAGES
Release to Public	Unclassified	24
	20. SECURITY CLASS *(This page)* Unclassified	22. PRICE

EPA Form 2220-1 (9-73)

THE DETERMINATION OF CHLORINATED HERBICIDES
IN INDUSTRIAL AND MUNICIPAL WASTEWATER

METHOD 615

1. Scope and Application

1.1 This method covers the determination of certain chlorinated herbicides. The following parent acids can be determined by this method:

Parameter	STORET No.	CAS No.
2,4-D	39736	94-75-7
Dalapon	--	75-99-0
2,4-DB	--	94-82-6
Dicamba	--	1918-00-9
Dichlorprop	--	120-36-5
Dinoseb	--	88-85-7
MCPA	--	94-74-6
MCPP	--	93-65-2
2,4,5-T	39740	93-76-5
2,4,5-TP	39760	93-72-1

1.2 This method is also applicable to the determination of salts and esters of these compounds. This includes, but is not limited to: the isobutyl and isooctyl esters of 2,4-D; the isobutyl and isooctyl esters of 2,4-DB; the isooctyl ester of MCPA; and the isooctyl ester of 2,4,5-TP. The actual form of each acid is not distinguished by this method. Results are calculated and reported for each listed parameter as total free acid.

1.3 This is a gas chromatographic (GC) method applicable to the determination of the compounds listed above in industrial and municipal discharges as provided under 40 CFR 136.1. Any modification of this method beyond those expressly permitted, shall be considered a major modification subject to application and approval of alternate test procedures under 40 CFR 136.4 and 136.5.

1.4 The method detection limit (MDL, defined in Section 15) for each parameter is listed in Table 1. The MDL for a specific wastewater may differ from those listed, depending upon the nature of interferences in the sample matrix.

1.5 This method is restricted to use by or under the supervision of analysts experienced in the use of gas chromatography and in the

interpretation of gas chromatograms. Each analyst must demonstrate the ability to generate acceptable results with this method using the procedure described in Section 8.2.

1.6 When this method is used to analyze unfamiliar samples for any or all of the compounds above, compound identifications should be supported by at least one additional qualitative technique. This method describes analytical conditions for alternative gas chromatographic columns that can be used to confirm measurements made with the primary column. Section 15 provides gas chromatograph/mass spectrometer (GC/MS) criteria appropriate for the qualitative confirmation of compound identifications.

2. Summary of Method

2.1 A measured volume of sample, approximately 1 liter, is acidified. The acid herbicides and their esters and salts are extracted with ethyl ether using a separatory funnel. The derivatives are hydrolyzed with potassium hydroxide and extraneous organic material is removed by a solvent wash. After acidification, the acids are exracted and converted to their methyl esters using diazomethane as the derivatizing agent. Excess reagent is removed, and the esters are determined by electron capture (EC) gas chromatography.[1]

3. Interferences

3.1 Method interferences may be caused by contaminants in solvents, reagents, glassware and other sample processing apparatus that lead to discrete artifacts or elevated baselines in gas chromatograms. All reagents and apparatus must be routinely demonstrated to be free from interferences under the conditions of the analysis by running laboratory reagent blanks as described in Section 8.5.

3.1.1 Glassware must be scrupulously cleaned.[2] Clean all glassware as soon as possible after use by thoroughly rinsing with the last solvent used in it. Follow by washing with hot water and detergent and thorough rinsing with dilute acid, tap and reagent water. Drain dry, and heat in an oven or muffle furnace at 400°C for 15 to 30 min. Do not heat volumetric ware. Thermally stable materials such as PCBs, might not be eliminated by this treatment. Thorough rinsing with acetone and pesticide quality hexane may be substituted for the heating. After drying and cooling, seal and store glassware in a clean environment to prevent any accumulation of dust or other contaminants. Store inverted or capped with aluminum foil.

3.1.2 The use of high purity reagents and solvents helps to minimize interference problems. Purification of solvents by distillation in all-glass systems may be required.

3.2 The acid forms of the herbicides are strong organic acids, which

react readily with alkaline substances and can be lost during analysis. Glassware and glass wool must be acid-rinsed with (1+9) hydrochloric acid and the sodium sulfate must be acidified with sulfuric acid prior to use to avoid this possibility.

3.3 Organic acids and phenols, especially chlorinated compounds, cause the most direct interference with the determination. Alkaline hydrolysis and subsequent extraction of the basic solution remove many chlorinated hydrocarbons and phthalate esters that might otherwise interfere with the electron capture analysis.

3.4 Matrix interferences may be caused by contaminants that are coextracted from the sample. The extent of matrix interferences will vary considerably from source to source, depending upon the nature and diversity of the industrial complex or municipality sampled. The cleanup procedure in Section 11 can be used to overcome many of these interferences, but unique samples may require additional cleanup approaches to achieve the MDL listed in Table 1.

4. Safety

4.1 The toxicity or carcinogenicity of each reagent used in this method has not been precisely defined; however, each chemical compound must be treated as a potential health hazard. From this viewpoint, exposure to these chemicals must be reduced to the lowest possible level by whatever means available. The laboratory is responsible for maintaining a current awareness file of OSHA regulations regarding the safe handling of the chemicals specified in this method. A reference file of material data handling sheets should also be made available to all personnel involved in the chemical analysis. Additional references to laboratory safety are available and have been identified [3-5] for the information of the analyst.

4.2 Diazomethane is a toxic carcinogen and can explode under certain conditions. The following precautions must be followed:

4.2.1 Use only a well ventilated hood - do not breath vapors.

4.2.2 Use a safety screen.

4.2.3 Use mechanical pipetting aides.

4.2.4 Do not heat above 90°C - EXPLOSION may result.

4.2.5 Avoid grinding surfaces, ground glass joints, sleeve bearings, glass stirrers - EXPLOSION may result.

4.2.6 Store away from alkali metals - EXPLOSION may result.

4.2.7 Solutions of diazomethane decompose rapidly in the presence of solid materials such as copper powder, calcium chloride,

and boiling chips.

5. Apparatus and Materials

5.1 Sampling equipment, for discrete or composite sampling.

5.1.1 Grab sample bottle - Amber borosilicate or flint glass, 1-liter or 1-quart volume, fitted with screw caps lined with TFE-fluorocarbon. Aluminum foil may be substituted for TFE if the sample is not corrosive. If amber bottles are not available, protect samples from light. The container and cap liner must be washed, rinsed with acetone or methylene chloride, and dried before use to minimize contamination.

5.1.2 Automatic sampler (optional) - Must incorporate glass sample containers for the collection of a minimum of 250 mL. Sample containers must be kept refrigerated at 4°C and protected from light during compositing. If the sampler uses a peristaltic pump, a minimum length of compressible silicone rubber tubing may be used. Before use, however, the compressible tubing must be thoroughly rinsed with methanol, followed by repeated rinsings with reagent water to minimize the potential for contamination of the sample. An integrating flow meter is required to collect flow proportional composites.

5.2 Glassware (All specifications are suggested. Catalog numbers are included for illustration only.)

5.2.1 Separatory funnels - 60-mL and 2000-mL, with TFE-fluorocarbon stopcocks, ground glass or TFE stoppers.

5.2.2 Concentrator tube, Kuderna-Danish - 10-mL, graduated (Kontes K-570050-1025 or equivalent). Calibration must be checked at the volumes employed in the test. Ground glass stopper is used to prevent evaporation of extracts.

5.2.3 Evaporative flask, Kuderna-Danish - 500-mL (Kontes K-570001-0500 or equivalent). Attach to concentrator tube with springs.

5.2.4 Snyder column, Kuderna-Danish - three-ball macro (Kontes K-503000-0121 or equivalent).

5.2.5 Snyder column, Kuderna-Danish - two-ball micro (Kontes K-569001-0219 or equivalent).

5.2.6 Erlenmeyer flask - Pyrex, 250 mL with 24/40 ground glass joint.

5.2.7 Vials - Amber glass, 10 to 15 mL capacity with TFE-fluorocarbon lined screw cap.

5.3 Boiling chips - approximately 10/40 mesh. Heat at 400°C for 30 min or Soxhlet extract with methylene chloride.

5.4 Water bath - Heated, with concentric ring cover, capable of temperature control (± 2°C). The bath should be used in a hood.

5.5 Balance - Analytical, capable of accurately weighing to the nearest 0.0001 g.

5.6 Diazomethane generator - assemble from two 20 x 150 mm test tubes, two Neoprene rubber stoppers and a source of nitrogen. The generator assembly is shown in Figure 1.

5.7 Glass wool - Acid washed (Supelco 2-0383 or equivalent).

5.8 Gas chromatograph - Analytical system complete with gas chromatograph suitable for on-column injection and all required accessories including syringes, analytical columns, gases, detector and strip-chart recorder. A data system is recommended for measuring peak areas.

5.8.1 Column 1 - 180 cm long x 4 mm ID glass, packed with 1.5% SP-2250/1.95% SP-2401 on Supelcoport (100/120 mesh) or equivalent. This column was used to develop the method performance statements in Section 16. Alternative columns may be used in accordance with the provisions described in Section 13.1.

5.8.2 Column 2 - 180 cm long x 4 mm ID glass, packed with 5% OV-210 on Gas Chrom Q (100/120 mesh) or equivalent.

5.8.3 Column 3 - 180 cm long x 2 mm ID glass, packed with 0.1% SP-1000 on Carbopak C (80/100 mesh) or equivalent.

5.8.3 Detector - Electron capture. This detector has proven effective in the analysis of wastewaters for the parameters listed in the scope and was used to develop the method performance statements in Section 15. Alternative detectors, including a mass spectrometer, may be used in accordance with the provisions described in Section 13.1.

6. Reagents

6.1 Reagent water - Reagent water is defined as a water in which an interferent is not observed at the method detection limit of each parameter of interest.

6.2 Acetone, hexane, methanol - Pesticide quality or equivalent.

6.3 Ethyl ether - Nanograde, redistilled in glass if necessary. Must be free of peroxides as indicated by EM Quant test strips. (Available from Scientific Products Co., Cat. No. P1126-8, and

other suppliers.) Procedures recommended for removal of peroxides are provided with the test strips. After cleanup, 20 mL ethyl alcohol preservative must be added to each liter of ether.

6.4 Sodium sulfate - (ACS) Granular, acidified, anhydrous. Heat treat in a shallow tray at 400°C for a minimum of 4 h to remove phthalates and other interfering organic substances. Alternatively, heat 16 h at 450-500°C in a shallow tray or Soxhlet extract with methylene chloride for 48 h. Acidify by slurrying 100 g sodium sulfate with enough ethyl ether to just cover the solid. Add 0.1 mL concentrated sulfuric acid and mix thoroughly. Remove the ether under vacuum. Mix 1 g of the resulting solid with 5 mL of reagent water and measure the pH of the mixture. It must be below pH 4. Store at 130°C.

6.5 Hydrochloric acid (1+9) - (ACS) Add one volume of concentrated acid to 9 volumes reagent water.

6.6 Potassium hydroxide solution - 37% aqueous solution (W:V). Dissolve 37 g ACS grade potassium hydroxide pellets in reagent water and dilute to 100 mL.

6.7 Sulfuric acid solution (1+1) - Slowly add 50 mL H_2SO_4 (sp. gr. 1.84) to 50 mL of reagent water.

6.8 Sulfuric acid solution (1+3) - Slowly add 25 mL H_2SO_4 (sp. gr. 1.84) to 75 mL of reagent water. Maintain at 4°C.

6.9 Carbitol - (Diethylene glycol monoethyl ether), ACS. Available from Aldrich Chemical Co.

6.10 Diazald - (N-methyl-N-nitroso-p-toluenesulfonamide), ACS. Available from Aldrich Chemical Co.

6.11 Silicic acid - Chromatographic grade, nominal 100 mesh. Store at 130°C.

6.12 Stock standard solutions (1.00 µg/µL) - Stock standard solutions can be prepared from pure standard materials or purchased as certified solutions.

6.12.1 Prepare stock standard solutions by accurately weighing about 0.0100 grams of pure acids. Dissolve the material in pesticide quality ethyl ether and dilute to volume in a 10-mL volumetric flask. Larger volumes can be used at the convenience of the analyst. If compound purity is certified at 96% or greater, the weight can be used without correction to calculate the concentration of the stock standard. Commercially prepared stock standards can be used at any concentration if they are certified by the manufacturer or by an independent source.

6.12.2 Transfer the stock standard solutions into Teflon-sealed screw-cap vials. Store at 4°C and protect from light. Stock standard solutions should be checked frequently for signs of degradation or evaporation, especially just prior to preparing calibration standards from them.

6.12.3 Stock standard solutions must be replaced after one week or sooner if comparison with check standards indicates a problem.

7. Calibration

7.1 Establish gas chromatographic operating parameters equivalent to those indicated in Table 1. The gas chromatographic system must be calibrated using the external standard technique.

7.2 External standard calibration procedure:

7.2.1 For each parameter of interest, prepare working standards of the free acids at a minimum of three concentration levels by adding accurately measured volumes of one or more stock standards to a 10-mL volumetric flask containing 1.0 mL methanol and diluting to volume with ethyl ether. One of the external standards should be representative of a concentration near, but above, the method detection limit. The other concentrations should correspond to the range of concentrations expected in the sample concentrates or should define the working range of the detector.

7.2.2 Prepare calibration standards by esterification of 1.00 mL volumes of the working standards as described in Section 11. Using injections of 2 to 5 µL of each calibration standard, tabulate peak height or area responses against the mass of free acid represented by the injection. The results can be used to prepare a calibration curve for each parameter. Alternatively, the ratio of the response to the mass injected, defined as the calibration factor ( ), can be calculated for each parameter at each standard concentration. If the relative standard deviation of the calibration factor is less than 10% over the working range, the average calibration factor can be used in place of a calibration curve.

7.2.3 The working calibration curve or calibration factor must be verified on each working shift by the preparation of one or more calibration standards. If the response for any parameter varies from the predicted response by more than ±10%, the test must be repeated using a fresh calibration standard. Alternatively, a new calibration curve or calibration factor must be prepared for that parameter.

7.3 Before using any cleanup procedure, the analyst must process a

series of calibration standards through the procedure to validate elution patterns and the absence of interference from the reagents.

8. Quality Control

8.1 Each laboratory using this method is required to operate a formal quality control program. The minimum requirements of this program consist of an initial demonstration of laboratory capability and the analysis of spiked samples as a continuing check on performance. The laboratory is required to maintain performance records to define the quality of data that is generated.

8.1.1 Before performing any analyses, the analyst must demonstrate the ability to generate acceptable accuracy and precision with this method. This ability is established as described in Section 8.2.

8.1.2 In recognition of the rapid advances occurring in chromatography, the analyst is permitted certain options to improve the separations or lower the cost of measurements. Each time such modifications to the method are made, the analyst is required to repeat the procedure in Section 8.2.

8.1.3 The laboratory must spike and analyze a minimum of 10% of all samples to monitor continuing laboratory performance. This procedure is described in Section 8.4.

8.2 To establish the ability to generate acceptable accuracy and precision, the analyst must perform the following operations.

8.2.1 Select a representative spike concentration for each compound (acid or ester) to be measured. Using stock standards, prepare a quality control check sample concentrate in acetone 1000 times more concentrated than the selected concentrations.

8.2.2 Using a pipet, add 1.00 mL of the check sample concentrate to each of a minimum of four 1000-mL aliquots of reagent water. A representative wastewater may be used in place of the reagent water, but one or more additional aliquots must be analyzed to determine background levels, and the spike level must exceed twice the background level for the test to be valid. Analyze the aliquots according to the method beginning in Section 10.

8.2.3 Calculate the average percent recovery (R), and the standard deviation of the percent recovery (s), for the results. Wastewater background corrections must be made before R and s calculations are performed.

8.2.4 Using the appropriate data from Table 2, determine the recovery and single operator precision expected for the

method, and compare these results to the values calculated in Section 8.2.3. If the data are not comparable, review potential problem areas and repeat the test.

8.3 The analyst must calculate method performance criteria and define the performance of the laboratory for each spike concentration and parameter being measured.

8.3.1 Calculate upper and lower control limits for method performance as follows:

Upper Control Limit (UCL) = R + 3 s

Lower Control Limit (LCL) = R - 3 s

where R and s are calculated as in Section 8.2.3. The UCL and LCL can be used to construct control charts[6] that are useful in observing trends in performance.

8.3.2 The laboratory must develop and maintain separate accuracy statements of laboratory performance for wastewater samples. An accuracy statement for the method is defined as R ± s. The accuracy statement should be developed by the analysis of four aliquots of wastewater as described in Section 8.2.2, followed by the calculation of R and s. Alternatively, the analyst may use four wastewater data points gathered through the requirement for continuing quality control in Section 8.4. The accuracy statements should be updated regularly.[6]

8.4 The laboratory is required to collect in duplicate a portion of their samples to monitor spike recoveries. The frequency of spiked sample analysis must be at least 10% of all samples or one spiked sample per month, whichever is greater. One aliquot of the sample must be spiked and analyzed as described in Section 8.2. If the recovery for a particular parameter does not fall within the control limits for method performance, the results reported for that parameter in all samples processed as part of the same set must be qualified as described in Section 14.3. The laboratory should monitor the frequency of data so qualified to ensure that it remains at or below 5%.

8.5 Before processing any samples, the analyst must demonstrate through the analysis of a 1-liter aliquot of reagent water that all glassware and reagents interferences are under control. Each time a set of samples is extracted or there is a change in reagents, a laboratory reagent blank must be processed as a safeguard against laboratory contamination.

8.6 It is recommended that the laboratory adopt additional quality assurance practices for use with this method. The specific practices that are most productive depend upon the needs of the laboratory and the nature of the samples. Field duplicates may be analyzed to monitor the precision of the sampling technique. When doubt exists over the identification of a peak on the chromatogram,

confirmatory techniques such as gas chromatography with a dissimilar column, specific element detector, or mass spectrometer must be used. Whenever possible, the laboratory should perform analysis of quality control materials and participate in relevant performance evaluation studies.

9. Sample Collection, Preservation, and Handling

9.1 Grab samples must be collected in glass containers. Conventional sampling practices[7] should be followed; however, the bottle must not be prerinsed with sample before collection. Composite samples should be collected in refrigerated glass containers in accordance with the requirements of the program. Automatic sampling equipment must be as free as possible of plastic and other potential sources of contamination.

9.2 The samples must be iced or refrigerated at 4°C from the time of collection until extraction.

9.3 All samples must be extracted within seven days and completely analyzed within 40 days of extraction.

10. Sample Extraction

10.1 Mark the water meniscus on the side of the sample bottle for later determination of sample volume. Pour the entire sample into a 2-liter separatory funnel. Check the pH with wide-range pH paper and adjust to pH less than 2 with sulfuric acid (1+1).

10.2 Add 150 mL ethyl ether to the sample bottle, cap the bottle and shake 30 s to rinse the walls. Transfer the solvent to the separatory funnel and extract the sample by shaking the funnel for 2 min with periodic venting to release excess pressure. Allow the organic layer to separate from the water phase for a minimum of 10 min. If the emulsion interface between layers is more than one third the volume of the solvent layer, the analyst must employ mechanical techniques to complete the phase separation. The optimum technique depends upon the sample, but may include stirring, filtration of the emulsion through glass wool, centrifugation or other physical means. Drain the aqueous phase into a 1000-mL Erlenmeyer flask and collect the extract in a 250-mL ground-glass Erlenmeyer flask containing 2 mL of 37% potassium hydroxide solution. Approximately 80 mL of the ethyl ether will remain dissolved in the aqueous phase.

10.3 Add a 50-mL volume of ethyl ether to the sample bottle and repeat the extraction a second time, combining the extracts in the Erlenmeyer flask. Perform a third extraction in the same manner.

10.4 Add 15 mL reagent water and 1 or 2 clean boiling chips to the 250-mL flask and attach a three-ball Snyder column. Prewet the Snyder column by adding 1 mL ethyl ether to the top. Place the

apparatus on a hot water bath (60 to 65°C), such that the bottom of the flask is bathed in the water vapor. Although the ethyl ether will evaporate in about 15 min, continue heating for a total of 60 min, beginning from the time the flask is placed on the water bath. Remove the apparatus and let stand at room temperature for at least 10 min.

10.5 Transfer the solution to a 60-mL separatory funnel using 5 to 10 mL of reagent water. Wash the basic solution twice by shaking for one min with 20-mL portions of ethyl ether. Discard the organic phase. The free acids remain in the aqueous phase.

10.6 Acidify the contents of the separatory funnel to pH 2 by adding 2 mL of cold (4°C) sulfuric acid (1+3). Test with pH indicator paper. Add 20 mL ethyl ether and shake vigorously for 2 min. Drain the aqueous layer into the 250-mL Erlenmeyer, then pour the organic layer into a 125-mL Erlenmeyer flask containing about 0.5 g of acidified anhydrous sodium sulfate. Repeat the extraction twice more with 10-mL aliquots of ethyl ether, combining all solvent in the 125-mL flask. Allow the extract to remain in contact with the sodium sulfate for approximately 2 h.

10.7 Assemble a Kuderna-Danish (K-D) concentrator by attaching a 10-mL concentrator tube to a 500-mL evaporative flask. Other concentration devices or techniques may be used in place of the K-D if the requirements of Section 8.2 are met.

10.8 Pour the combined extract through a funnel plugged with acid washed glass wool, and collect the extract in the K-D in concentrator. Use a glass rod to crush any caked sodium sulfate during the transfer. Rinse the Erlenmeyer flask and column with 20 to 30 mL of ethyl ether to complete the quantitative transfer.

10.9 Add 1 to 2 clean boiling chips to the evaporative flask and attach a three-ball Snyder column. Prewet the Snyder column by adding about 1 mL ethyl ether to the top. Place the K-D apparatus on a hot water bath, 60 to 65°C, so that the concentrator tube is partially immersed in the hot water, and the entire lower rounded surface of the flask is bathed with hot vapor. Adjust the vertical position of the apparatus and the water temperature as required to complete the concentration in 15 to 20 min. At the proper rate of distillation the balls of the column will actively chatter but the chambers will not flood. When the apparent volume of liquid reaches 1 mL, remove the K-D apparatus and allow it to drain and cool for at least 10 min.

10.10 Remove the Snyder column and rinse the flask and its lower joint into the concentrator tube with 1 to 2 mL of ethyl ether. A 5-mL syringe is recommended for this operation. Add a fresh boiling chip. Attach a micro-Snyder column to the concentrator tube and prewet the column by adding about 0.5 mL of ethyl ether to the top. Place the micro K-D apparatus on the water bath so that the

concentrator tube is partially immersed in the hot water. Adjust the vertical position of the apparatus and the water temperature as required to complete concentration in 5 to 10 min. When the apparent volume of liquid reaches 0.5 mL, remove the micro K-D from the bath and allow it to drain and cool. Remove the micro Snyder column and add 0.1 mL of methanol. Rinse the walls of the concentrator tube while adjusting the volume to 1.0 mL with ethyl ether.

11. Esterification of Acids

11.1 Assemble the diazomethane generator (See Figure 1) in a hood using two 20 x 150 mm test tubes. Use neoprene rubber stoppers with holes drilled in them to accommodate glass delivery tubes. The exit tube must be drawn to a point to bubble diazomethane through the sample extract.

11.2 Add 5 mL of ethyl ether to the first test tube. Add 1 mL of ethyl ether, 1 mL of carbitol, 1.5 mL of 37% aqueous KOH, and 0.1 to 0.2 g Diazald to the second test tube. Immediately place the exit tube into the concentrator tube containing the sample extract. Apply nitrogen flow (10 mL/min) to bubble diazomethane through the extract for 10 min or until the yellow color of diazomethane persists.

11.3 Remove the concentrator tube and seal it with a Neoprene or Teflon stopper. Store at room temperature in a hood for 20 min.

11.4 Destroy any unreacted diazomethane by adding 0.1 to 0.2 g silicic acid to the concentrator tube. Allow to stand until the evolution of nitrogen gas has stopped. Adjust the sample volume to 10.0 mL with hexane. Stopper the concentrator tube and store refrigerated if further processing will not be performed immediately. It is recommended that the methylated extracts be analyzed immediately to minimize any transesterification and other potential reactions that may occur. Analyze by gas chromatography.

11.5 Determine the original sample volume by refilling the sample bottle to the mark and transferring the water to a 1000-mL graduated cylinder. Record the sample volume to the nearest 5 mL.

12. Cleanup and Separation

12.1 No cleanup procedures were required to analyze the wastewaters described in Section 16. If particular circumstances demand the use of a cleanup procedure, the analyst must determine the elution profile and demonstrate that the recovery of each compound of interest for the cleanup procedure is no less than 85%.

13. Gas Chromatography

13.1 Table 1 summarizes the recommended operating conditions for the

gas chromatograph. Included in this table are estimated retention times and method detection limits that can be achieved by this method. Examples of the separations achieved for the methyl esters are shown in Figures 2 to 4. Other packed columns, chromatographic conditions, or detectors may be used if the requirements of Section 8.2 are met. Capillary (open-tubular) columns may also be used if the relative standard deviations of responses for replicate injections are demonstrated to be less than 6% and the requirements of Section 8.2 are met.

13.2 Calibrate the system daily as described in Section 7.

13.3 Inject 1 to 5 µL of the sample extract using the solvent-flush technique.[8] Record the volume injected to the nearest 0.05 µL, and the resulting peak size in area or peak height units. An automated system that consistently injects a constant volume of extract may also be used.

13.4 The width of the retention time window used to make identifications should be based upon measurements of actual retention time variations of standards over the course of a day. Three times the standard deviation of a retention time can be used to calculate a suggested window size for a compound. However, the experience of the analyst should weigh heavily in the interpretation of chromatograms.

13.5 If the response for the peak exceeds the working range of the system, dilute the extract and reanalyze.

13.6 If the measurement of the peak response is prevented by the presence of interferences, further cleanup is required.

14. Calculations

14.1 Determine the concentration of individual compounds in the sample. Calculate the amount of free acid injected from the peak response using the calibration curve or calibration factor in Section 7.2.2. The concentration in the sample can be calculated as follows:

$$\text{Concentration, } \mu g/L = \frac{(A)(V_t)}{(V_i)(V_s)}$$

where:

A = Amount of material injected, in nanograms.
V_i = Volume of extract injected in µL.
V_t = Volume of total extract in µL.
V_s = Volume of water extracted in mL.

14.2 Report results in micrograms per liter as acid equivalent without correction for recovery data. When duplicate and spiked samples

are analyzed, report all data obtained with the sample results.

14.3 For samples processed as part of a set where the laboratory spiked sample recovery falls outside of the control limits in Section 8.3, data for the affected parameters must be labeled as suspect.

15. GC/MS Confirmation

15.1 It is recommended that GC/MS techniques be judiciously employed to support qualitative compound identifications made with this method. The mass spectrometer should be capable of scanning the mass range from 35 amu to a mass 50 amu above the molecular weight of the methyl ester of the acid herbicide. The instrument must be capable of scanning the mass range at a rate to produce at least 5 scans per peak but not to exceed 7 s per scan utilizing a 70 V (nominal) electron energy in the electron impact ionization mode. A GC to MS interface constructed of all-glass or glass-lined materials is recommended. A computer system should be interfaced to the mass spectrometer that allows the continuous acquisition and storage on machine readable media of all mass spectra obtained throughout the duration of the chromatographic program.

15.2 Gas chromatographic columns and conditions should be selected for optimum separation and performance. The conditions selected must be compatible with standard GC/MS operating practices. Chromatographic tailing factors of less than 5.0 must be achieved.[9]

15.3 At the beginning of each day that confirmatory analyses are to be performed, the GC/MS system must be checked to see that all decafluorotriphenyl phosphine (DFTPP) performance criteria are achieved.[10]

15.4 To confirm an identification of a compound, the background corrected mass spectrum of the methyl ester must be obtained from the sample extract and compared with a mass spectrum from a stock or calibration standard analyzed under the same chromatographic conditions. It is recommended that at least 25 nanograms of material be injected into the GC/MS. The criteria below must be met for qualitative confirmation.

15.4.1 All ions that are present above 10% relative abundance in the mass spectrum of the standard must be present in the mass spectrum of the sample with agreement to plus or minus 10%. For example, if the relative abundance of an ion is 30% in the mass spectrum of the standard, the allowable limits for the relative abundance of that ion in the mass spectrum for the sample would be 20% to 40%.

15.4.2 The retention time of the compound in the sample must be within 6 seconds of the same compound in the standard solution.

15.4.3 Compounds that have very similar mass spectra can be explicitly identified by GC/MS only on the basis of retention time data.

15.5 Where available, chemical ionization mass spectra may be employed to aid in the qualitative identification process.

15.6 Should these MS procedures fail to provide satisfactory results, additional steps may be taken before reanalysis. These may include the use of alternate packed or capillary GC columns or additional cleanup.

16. Method Performance

16.1 The method detection limit (MDL) is defined as the minimum concentration of a substance that can be measured and reported with 99% confidence that the value is above zero.[11] The MDL concentrations listed in Table 1 were obtained from reagent water with an electron capture detector.[1]

16.2 In a single laboratory (West Coast Technical Services, Inc.), using reagent water and effluents from publicly owned treatment works (POTW), the average recoveries presented in Table 2 were obtained.[1] The standard deviations of the percent recoveries of these measurements are also included in Table 2.

References

1. "Pesticide Methods Evaluation," Letter Report #33 for EPA Contract No. 68-03-2697. Available from U.S. Environmental Protection Agency, Environmental Monitoring and Support Laboratory, Cincinnati, Ohio 45268.

2. ASTM Annual Book of Standards, Part 31, D3694, "Standard Practice for Preparation of Sample Containers and for Preservation, " American Society for Testing and Materials, Philadelphia, PA, p. 679, 1980.

3. "Carcinogens - Working with Carcinogens," Department of Health, Education, and Welfare, Public Health Service, Center for Disease Control, National Institute for Occupational Safety and Health, Publication No. 77-206, Aug. 1977.

4. "OSHA Safety and Health Standards, General Industry," (29 CFR 1910), Occupational Safety and Health Administration, OSHA 2206, (Revised, January 1976).

5. "Safety in Academic Chemistry Laboratories," American Chemical Society Publication, Committee on Chemical Safety, 3rd Edition, 1979.

6. "Handbook for Analytical Quality Control in Water and Wastewater Laboratories," EPA-600/4-79-019, U. S. Environmental Protection Agency, Environmental Monitoring and Support Laboratory - Cincinnati, Ohio 45268, March 1979.

7. ASTM Annual Book of Standards, Part 31, D3370, "Standard Practice for Sampling Water," American Society for Testing and Materials, Philadelphia, PA, p. 76, 1980.

8. Burke, J. A., "Gas Chromatography for Pesticide Residue Analysis; Some Practical Aspects," Journal of the Association of Official Analytical Chemists, 48, 1037 (1965).

9. McNair, H.M. and Bonelli, E. J., "Basic Chromatography," Consolidated Printing, Berkeley, California, p. 52, 1969.

10. Eichelberger, J.W., Harris, L.E., and Budde, W.L. "Reference Compound to Calibrate Ion Abundance Measurement in Gas Chromatography-Mass Spectrometry," Analytical Chemistry, 47, 995 (1975).

11. Glaser, J.A. et.al, "Trace Analysis for Wastewaters," Environmental Science & Technology, 15, 1426 (1981).

TABLE 1

CHROMATOGRAPHIC CONDITIONS AND METHOD DETECTION LIMITS

Parameter (as methyl ester)	Retention Time (min.) Column 1	Column 2	Column 3	Method Detection Limit µg/L
Dicamba	1.2	1.0	--	0.27
2,4-D	2.0	1.6	--	1.2
2,4,5-TP	2.7	2.0	--	0.17
2,4,5-T	3.4	2.4	--	0.20
2,4-DB	4.1	--	--	0.91
Dalapon	--	--	5.0	5.8
MCPP	3.4	--	--	192
MCPA	4.1	--	--	249
Dichlorprop	4.8	--	--	0.65
Dinoseb	11.2	--	--	0.07

Column 1 conditions: Supelcoport (100/120 mesh) coated with 1.5% SP-2250/ 1.95% SP-2401 packed in a 1.8 m long x 4 mm ID glass column with 95% argon/5% methane carrier gas at a flow rate of 70 mL/min. Column temperature: isothermal at 185°C, except for MCPP, MCPA, dichlorprop and dinoseb, where the column temperature was held at 140°C for 6 min and then programmed to 200°C at 10°/min. An electron capture detector was used to measure MDL.

Column 2 conditions: Gas Chrom Q (100/120 mesh) coated with 5% OV-210 packed in a 1.8 m long x 4 mm ID glass column with 95% argon/5% methane carrier gas at a flow rate of 70 mL/min. Column temperature: isothermal at 185°C.

Column 3 conditions: Carbopak C (80/100 mesh) coated with 0.1% SP-1000 packed in a 1.8 m long x 2 mm ID glass column with nitrogen carrier gas at a flow rate of 25 mL/min. Column temperature: programmed at injection from 100°C to 150°C at 10°/min.

TABLE 2

SINGLE OPERATOR ACCURACY AND PRECISION*

Parameter	Sample Type	Spike (µg/L)	Mean Recovery (%)	Standard Deviation (%)
2,4-D	DW	10.9	75	4
	MW	10.1	77	4
	MW	200	65	5
Dalapon	DW	23.4	66	8
	MW	23.4	96	13
	MW	468	81	9
2,4-DB	DW	10.3	93	3
	MW	10.4	93	3
	MW	208	77	6
Dicamba	DW	1.2	79	7
	MW	1.1	86	9
	MW	22.2	82	6
Dichlorprop	DW	10.7	97	2
	MW	10.7	72	3
	MW	213	100	2
Dinoseb	MW	0.5	86	4
	MW	102	81	3
MCPA	DW	2020	98	4
	MW	2020	73	3
	MW	21400	97	2
MCPP	DW	2080	94	4
	MW	2100	97	3
	MW	20440	95	2
2,4,5-T	DW	1.1	85	6
	MW	1.3	83	4
	MW	25.5	78	5
2,4,5-TP	DW	1.0	88	5
	MW	1.3	88	4
	MW	25.0	72	5

*All results based upon seven replicate analyses.

DW = Reagent water
MW = Municipal water

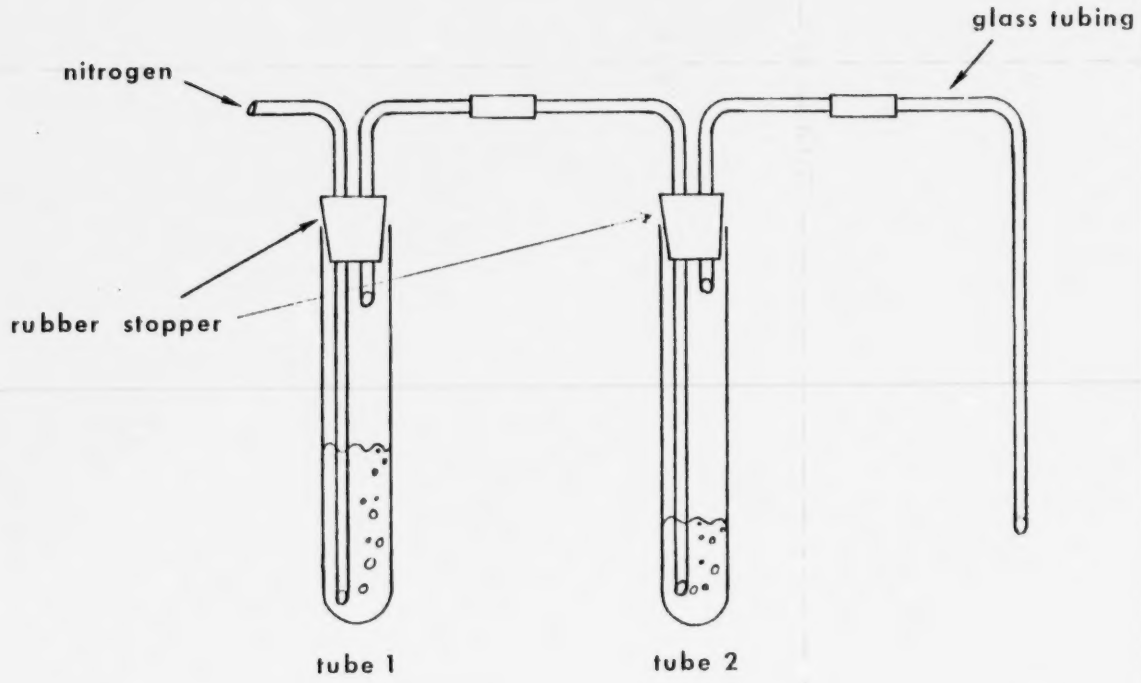


Figure 1. Diazomethane generator.

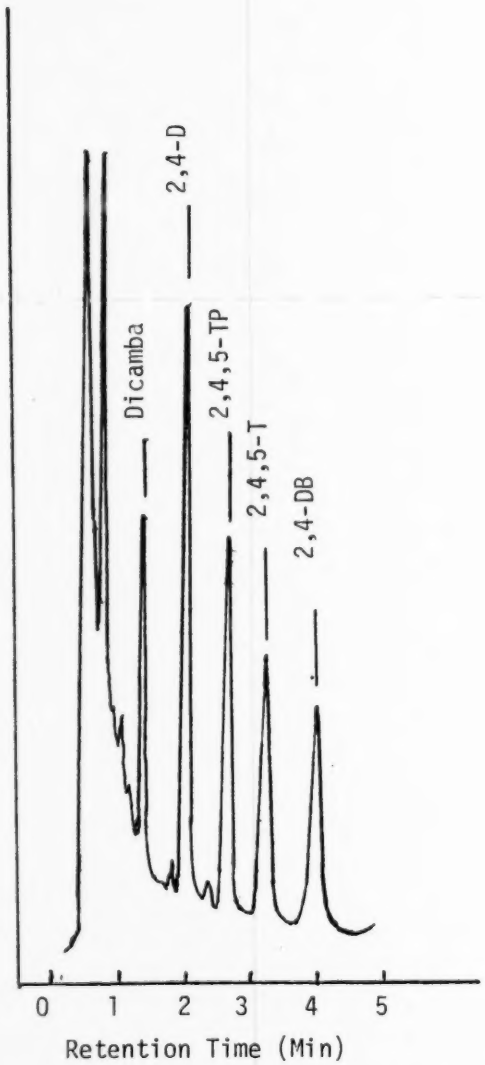


Figure 2. Gas chromatogram of methyl esters of chlorinated herbicides on Column 1. For conditions, see Table 1.

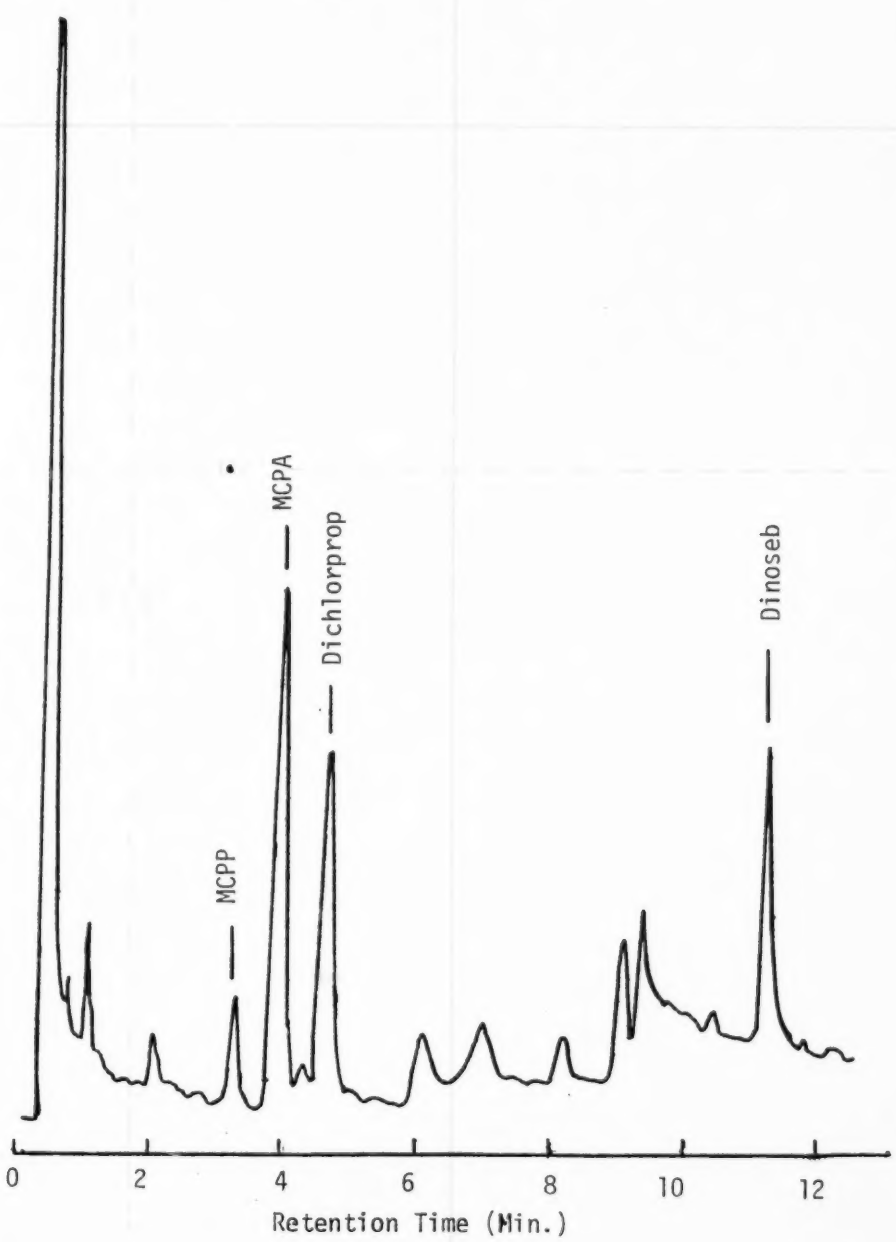


Figure 3. Gas chromatogram of methyl esters of chlorinated herbicides on Column 1. For conditions, see Table 1.

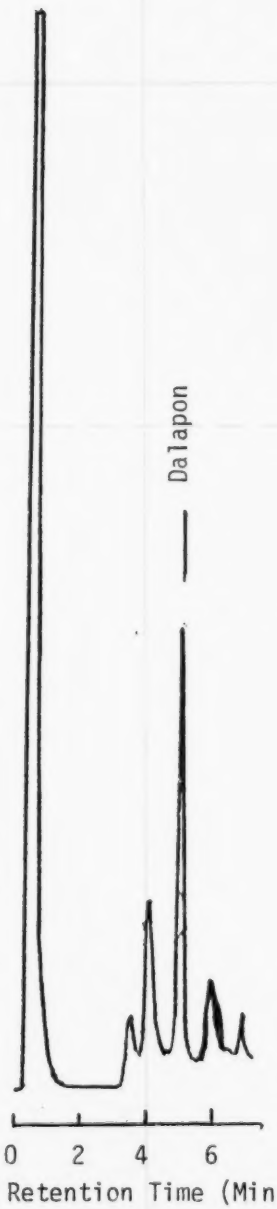


Figure 4. Gas chromatogram of methyl ester of dalapon on Column 3. For conditions, see Table 1.